Dkt. 4396 CA-PCT-US/JPW/SHS

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants : Graham P. Allaway, et al.

Serial No. : Not Yet Known (continuation of U.S. Serial No 08/973,601, filed March 16, 1998)

Filed : October 5, 1999

For METHODS FOR USING RESONANCE ENERGY TRANSFER-BASED ASSAY OF HIV-1 ENVELOPE GLYCOPROTEIN-MEDIATED MEMBRANE FUSION, AND KITS FOR PRACTICING SAME

1185 Avenue of the Americas
New York, New York 10036
October 5, 1999

Assistant Commissioner for Patents
Washington, D.C. 20231

Copy App. #2

Sir:

PRELIMINARY AMENDMENT

Please amend the subject application as follows:

In the specification:

On page 1, line 6, after the words "This application is a" and before the words "continuation-in-part" please insert the following words: --continuation of United States Serial No. 08/973,601, filed March 16, 1998, which is a national stage application, filed under 35 U.S.C. §371 of PCT/US96/09894, filed June 7, 1996, which is a--.

On page 1, line 7, before the words "Serial No." and after the words "filed June 7" please delete "08/175,515," and insert the following --08/475,515,--.

In the claims:

Please cancel claims 1-6 without prejudice to applicants' right to pursue the subject matter of these claims in a later-filed application and add the following new claims:

--7. (New) An agent determined to be capable of specifically inhibiting the fusion of a macrophage-tropic primary isolate of HIV-1 to a $CD4^+$ cell, but not a T cell-tropic isolate of HIV-1 to a $CD4^-$ cell, using a method which comprises:

(a) contacting (i) a first appropriate $CD4^+$ cell, which is labeled with a first dye, with (ii) a cell expressing the HIV-1 envelope glycoprotein of the macrophage-tropic primary isolate of HIV-1 on its surface, which is labeled with a second dye, in the presence of an excess of the agent under conditions which would normally permit the fusion of the $CD4^+$ cell to the cell expressing the HIV-1 envelope glycoprotein on its surface in the absence of the agent, the first and second dyes being selected so as to allow resonance energy transfer between the dyes;

(b) exposing the product of step (a) to conditions which would result in resonance energy transfer if fusion has occurred; and

(c) determining whether there is a reduction of resonance energy transfer, when compared with the resonance energy transfer in the absence of the agent;

(d) contacting (i) a second appropriate $CD4^+$ cell, which is labeled with a first dye, with (ii) a cell expressing the HIV-1 envelope glycoprotein of a T cell-tropic isolate of HIV-1 on its surface, which is labeled with a second dye, in the presence of an excess of the agent under conditions which would normally permit the fusion of the $CD4^+$ cell to the cell expressing the HIV-1 envelope glycoprotein on its surface in the absence of the agent, the first and second dyes being selected so as to allow resonance energy

transfer between the dyes;

(e) exposing the product of step (d) to conditions which would result in resonance energy transfer if fusion has occurred; and

(f) determining whether there is a reduction of resonance energy transfer, when compared with the resonance energy transfer in the absence of the agent, wherein a decrease in transfer in step (c) but not step (f) indicates that the agent is capable of specifically inhibiting fusion of the macrophage-tropic primary isolate of HIV-1 to $CD4^+$ cells and a decrease in transfer in step (f) but not step (c) indicates that the agent is capable of specifically inhibiting the fusion of a macrophage-tropic primary isolate of HIV-1 to the $CD4^+$ cells.--

--8. (New) The agent of claim 7, wherein the agent is an antibody.--

--9. (New) An agent capable of specifically inhibiting the fusion of a macrophage tropic primary isolate of HIV-1 with a CD+ cell susceptible to infection by a macrophage-tropic primary isolate of HIV-1.--

--10. (New) A method of inhibiting fusion of a macrophage-tropic primary isolate of HIV-1 with a CD+ cell susceptible to infection by a macrophage-tropic primary isolate of HIV-1 which comprises contacting the $CD4^+$ cell with an amount of an agent capable of specifically inhibiting such fusion so as to thereby inhibit such fusion.--

--11. (New) An agent determined to be capable of specifically inhibiting the fusion of a T cell-tropic isolate

of HIV-1 to a $CD4^+$ cell, but not a macrophage-tropic primary isolate of HIV-1 to a CD4+ cell, using a method which comprises:

(a) contacting (i) a first appropriate $CD4^+$ cell, which is labeled with a first dye, with (ii) a cell expressing the HIV-1 envelope glycoprotein of the macrophage-tropic primary isolate of HIV-1 on its surface, which is labeled with a second dye, in the presence of an excess of the agent under conditions which would normally permit the fusion of the $CD4^+$ cell to the cell expressing the HIV-1 envelope glycoprotein on its surface in the absence of the agent, the first and second dyes being selected so as to allow resonance energy transfer between the dyes;

(b) exposing the product of step (a) to conditions which would result in resonance energy transfer if fusion has occurred; and

(c) determining whether there is a reduction of resonance energy transfer, when compared with the resonance energy transfer in the absence of the agent;

(d) contacting (i) a second appropriate $CD4^+$ cell, which is labeled with a first dye, with (ii) a cell expressing the HIV-1 envelope glycoprotein of a T cell-tropic isolate of HIV-1 on its surface, which is labeled with a second dye, in the presence of an excess of the agent under conditions which would normally permit the fusion of the $CD4^+$ cell to the cell expressing the HIV-1 envelope glycoprotein on its surface in the absence of the agent, the first and second dyes being selected so as to allow resonance energy transfer between the dyes;

(e) exposing the product of step (d) to conditions

which would result in resonance energy transfer if fusion has occurred; and

(f) determining whether there is a reduction of resonance energy transfer, when compared with the resonance energy transfer in the absence of the agent, wherein a decrease in transfer in step (c) but not step (f) indicates that the agent is capable of specifically inhibiting fusion of the macrophage-tropic primary isolate of HIV-1 to $CD4^+$ cells and a decrease in transfer in step (f) but not step (c) indicates that the agent is capable of specifically inhibiting the fusion of a T cell-tropic isolate of HIV-1 to the $CD4^+$ cells.--

--12. (New) The agent of claim 11, wherein the agent is an antibody.--

--13. (New) An agent capable of specifically inhibiting the fusion of a T cell-tropic isolate of HIV-1 with a $CD4^+$ cell susceptible to infection by a T cell-tropic isolate of HIV-1.--

--14. (New) A method of inhibiting fusion of a T cell-tropic isolate of HIV-1 with a $CD4^+$ cell susceptible to infection by a T cell-tropic isolate of HIV-1 which comprises contacting the $CD4^+$ cell with an amount of an agent capable of specifically inhibiting such fusion so as to thereby inhibit such fusion.--

In the Abstract:

Please add page 67 containing the abstract of the disclosure, a copy of which is attached hereto as **Exhibit A**.